Translation

PCT/EP2003/012859

PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference U30034PCT	FOR FURTHER ACTION	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
International application No. PCT/EP2003/012859	International filing date (*day/month/year*) 17 November 2003 (17.11.2003)	Priority date (*day/month/year*) 16 November 2002 (16.11.2002)
International Patent Classification (IPC) or national classification and IPC G01N 33/50		
Applicant JOHANN WOLFGANG GOETHE-UNIVERSITÄT FRANKFURT AM MAIN		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 8 sheets, including this cover sheet.

☐ This report is also accompanied by ANNEXES, i.e., sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

These annexes consist of a total of _____ sheets.

3. This report contains indications relating to the following items:

- I ☒ Basis of the report
- II ☐ Priority
- III ☒ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- IV ☐ Lack of unity of invention
- V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- VI ☐ Certain documents cited
- VII ☐ Certain defects in the international application
- VIII ☐ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
25 May 2004 (25.05.2004)	10 September 2004 (10.09.2004)
Name and mailing address of the IPEA/EP Facsimile No.	Authorized officer Telephone No.

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No.
PCT/EP2003/012859

I. Basis of the report

1. With regard to the **elements** of the international application:*

☐ the international application as originally filed

☒ the description:

pages 1-23, as originally filed
pages _____, filed with the demand
pages _____, filed with the letter of _____

☒ the claims:

pages 1-24, as originally filed
pages _____, as amended (together with any statement under Article 19
pages _____, filed with the demand
pages _____, filed with the letter of _____

☒ the drawings:

pages 1/4-4/4, as originally filed
pages _____, filed with the demand
pages _____, filed with the letter of _____

☐ the sequence listing part of the description:

pages _____, as originally filed
pages _____, filed with the demand
pages _____, filed with the letter of _____

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.
These elements were available or furnished to this Authority in the following language _____ which is:

☐ the language of a translation furnished for the purposes of international search (under Rule 23.1(b)).
☐ the language of publication of the international application (under Rule 48.3(b)).
☐ the language of the translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

☐ contained in the international application in written form.
☐ filed together with the international application in computer readable form.
☐ furnished subsequently to this Authority in written form.
☐ furnished subsequently to this Authority in computer readable form.
☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. ☐ The amendments have resulted in the cancellation of:

☐ the description, pages _____
☐ the claims, Nos. _____
☐ the drawings, sheets/fig _____

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).**

* *Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rule 70.16 and 70.17).*

** *Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.*

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No.
PCT/EP2003/012859

III. Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

1. The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non obvious), or to be industrially applicable have not been examined in respect of:

☐ the entire international application.

☐ claims Nos. _____

because:

☒ the said international application, or the said claims Nos. 1-21 and 23-24 relate to the following subject matter which does not require an international preliminary examination *(specify)*:

See Supplemental sheets

☐ the description, claims or drawings *(indicate particular elements below)* or said claims Nos. _____ are so unclear that no meaningful opinion could be formed *(specify)*:

☐ the claims, or said claims Nos. _____ are so inadequately supported by the description that no meaningful opinion could be formed.

☐ no international search report has been established for said claims Nos. _____.

2. A meaningful international preliminary examination cannot be carried out due to the failure of the nucleotide and/or amino acid sequence listing to comply with the standard provided for in Annex C of the Administrative Instructions:

☐ the written form has not been furnished or does not comply with the standard.

☐ the computer readable form has not been furnished or does not comply with the standard.

Supplemental Box
(To be used when the space in any of the preceding boxes is not sufficient)

Continuation of: Box III.1

Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

Claims 1 to 21, 23 and 24 relate to a method which is carried out on a living body ("obtaining a sample"). The subject matter thus falls under PCT Rule 67.1 (iv), and consequently no expert opinion has been established with regard to the industrial applicability of these claims (PCT Article 34(4)(a)(i)).

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Claims	22	YES
	Claims	1-21, 23-24	NO
Inventive step (IS)	Claims		YES
	Claims	1-24	NO
Industrial applicability (IA)	Claims	22	YES
	Claims		NO

2. Citations and explanations

1. Citations

Reference is made to the following documents:

D1: ROESSIG L et al.: "Evidence for increased circulating apoptotic endothelial cells in patients with coronary artery disease", EUROPEAN HEART JOURNAL, Vol. 23, No. Abstract Supplement, page 656, XP009028084, Congress of the European Society of Cardiology; Berlin, Germany; August 31 - September 04, 2002, ISSN 0195-668X (ISSN print)

D2: ROSSIG LOTHAR et al.: "Levels of circulating apoptotic endothelial cells reflect disease activity in patients with coronary artery disease", CIRCULATION, Vol. 106, No. 19, Supplement, 5 November 2002 (2002-11-05), pages II-U, XP009028097, Abstracts from Scientific Sessions; Chicago, IL, USA; November 17-20, 2002, ISSN 0009-7322 (ISSN print)

D3: GEORGE F et al.: "Cytofluorometric detection of human endothelial cells in whole blood using S-Endo 1 monoclonal antibody", JOURNAL OF IMMUNOLOGICAL METHODS 1991 NETHERLANDS, Vol. 139, No. 1, 1991, pages 65-75, XP009027334, ISSN 0022-1759 (cited in the application)

D4: MUTIN MURIELLE et al.: "Direct evidence of endothelial injury in acute myocardial infarction and unstable angina by demonstration of circulating endothelial cells", BLOOD, Vol. 93, No. 9, 1 May 1999 (1999-05-01), pages 2951-2958, XP001180171, ISSN 0006-4971 (cited in the application)

D5: VASA MARIUCA et al.: "Statin therapy increases the number and stimulates migration of endothelial progenitor cells in patients with stable coronary artery disease and acute myocardial infarction", CIRCULATION, Vol. 104, No. 17, Supplement, 23 October 2001 (2001-10-23), pages II-725, XP002274963, Scientific Sessions 2001 of the American Heart Association; Anaheim, California, USA; November 11-14, 2001, ISSN 0009-7322

D6: MONESTIROLI SILVIA et al.: "Kinetics and viability of circulating endothelial cells as surrogate angiogenesis marker in an animal model of human lymphoma", CANCER RESEARCH, Vol. 61, No. 11, 1 June 2001 (2001-06-01), pages 4341-4344, XP002274899, ISSN 0008-5472

D7: MANCUSO PATRIZIA et al.: "Resting and activated endothelial cells are increased in the peripheral blood of cancer patients", BLOOD, Vol. 97, No. 11, 1 June 2001 (2001-06-01), pages 3658-3661, XP002274898, ISSN 0006-4971 (cited in the application)

D8: DIGNAT-GEORGE FRANCOISE et al.: "Circulating endothelial cells in acute coronary syndromes", BLOOD, Vol. 95, No. 2, 15 January 2000 (2000-01-15), page 728, XP009028120, ISSN 0006-4971

2. Novelty, inventive step and industrial applicability (PCT Article 33)

2.1 Document D1 describes a method for detecting endothelial cells associated with cardiovascular diseases in blood samples. Endothelial cell markers (CD146 and von Willebrand factor), apoptosis markers (annexin V) and endothelial precursor cell markers (CD133) are used (see the abstract), and a non-endothelial cell marker is also detected (CD45). Claims 1 to 5 and 7 to 21 of the present application are thus anticipated by D1. The subject matter of claim 6 (use of blood samples with added coagulation inhibitor) also appears to be implicit in D1. **The subject matter of claims 1 to 21 therefore lacks novelty.**

Document D2 also describes a similar method in which circulating apoptotic endothelial cells are detected. The features of claims 1 to 21 appear to be either disclosed by or implicit in D2. For the sake of completeness it is noted that the aforementioned method is also anticipated by documents D3, D4 and D5. **The subject matter of claims 1 to 21 therefore lacks novelty.**

Document D4 describes the aforementioned method in the investigation of acute myocardial infarction and its usefulness in the monitoring of treatment for vascular diseases (pages 215 and 219). **Claim 23 therefore appears to lack novelty in relation to D4.**

Document D5 also describes the use of a similar method (in this case the measuring of CD34-positive floating endothelial cells) during statin therapy for patients with coronary artery disease. The features of claims 23 and 24 are all either disclosed by or implicit in D5. **The subject matter of claims 23 and 24 therefore lacks novelty in relation to D5.**

The following feature (claim 22) is not disclosed in the prior art (documents D1 to D8):

- A diagnostic kit for carrying out the method according to one of claims 1 to 21.

Summary:
The subject matter of claims 1 to 21, 23 and 24 lacks novelty; claim 22 appears to be novel.

2.2 The subject matter of claims 1 to 21, 23 and 24 is already known (see above) and therefore does not involve an inventive step.

It is noted that even if the claims were to be amended in such a way as to make them novel (for example, by limiting them to features which are not disclosed in the same combination in documents D1 to D5), they would probably

still not involve an inventive step. All the features mentioned in the description and in the claims are already known from documents D1 to D6 or appear to be conventional measures. For a person skilled in the art, incorporating these measures would be an obvious and routine procedure for modifying the known method. The features in question do not appear to be suitable for establishing an inventive step.

2.3 As indicated above, the subject matter of claim 22 is novel over documents D1 to D5, since the term "kit" is not explicitly mentioned in any of these documents. However, the packing of test components into a box cannot be considered inventive if it is well known that these components are used in conjunction with each other (see documents D1 to D8). **Claim 22 therefore fails to meet the PCT requirement of inventive step.**

2.4 Claim 22 appears to be industrially applicable.

3. Further objections

3.1 Claim 22 is not clear because it merely refers to other claims and does not contain any technical features of its own. In order to meet the requirement of clarity (PCT Article 6), all the components of the claimed test kit should be specified in the claim.

3.2 It is clear from the application that the problem addressed by the application is that of providing a method for detecting shed floating endothelial cells and/or endothelial precursor cells in order to detect changes in the endothelial function associated with cardiovascular diseases (see page 5). Certain essential features of the invention do not appear to be specified in the claims (for example, the fact that floating cells are detected). The applicant is reminded that an independent claim must contain all the technical features that are essential to the definition of the invention (PCT Article 6 in conjunction with PCT Rule 6.3(b)).

3.3 Some of the citations in the description are either incomplete (page 4: Dignat-George et al.) or wrong (pages 7 and 19: Vasa et al.) and should be corrected.

3.4 Contrary to the requirements of PCT Rule 5.1(a)(ii), the description does not cite documents D1, D2, D5 and D8 or give an account of the relevant prior art disclosed therein.